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RESEARCH OPINIONS IN ANIMAL & VETERINARY SCIENCES

Effects of dietary L-carnitine supplementation on testicular histology of Iraqi drakes

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Abstract

The present study was conducted to determine the effects of dietary L-carnitine at different levels on testicular histology in Iraqi drakes. Forty-eight, 30-wk-old Iraqi drakes, were fed the same basal diet that was supplemented with 0 (control), 50, 100 or 150 mg of L-carnitine/kg of diet for 12 weeks. The supplementation of dietary L-carnitine at levels of 50, 100 or 100 mg/kg to a basal diet significantly increased absolute and relative weight of testis, germinal cell thickness, seminiferous tubules lumen diameter and seminiferous tubules diameter. However, supplementation of the drake ration with different levels of L-carnitine significantly increased volume density of spermatocytes, spermatids, sperms, spermatogenic cells, Sertoli cells, total seminiferous tubules, Leydig cells, blood vessels and total interstitium and significantly decreased volume density of vacuoles, lumen, interstitial spaces and total seminiferous tubules/total interstitium ratio. Moreover, feeding diets containing different levels of L-carnitine resulted in significant increase in relative weight of spermatogonia, spermatocytes, spermatids, sperms, spermatogenic cells, Sertoli cells, total seminiferous tubules, myoid cells, Leydig cells, blood vessels and total interstitium and significant decrease in relative weight of total seminiferous tubules/total interstitium. These results suggest that dietary L-carnitine supplementation significantly improve testes histological traits of drakes. Therefore, L-carnitine can be used as beneficial tool for improving reproductive performance of male birds.

Keywords: Carnitine; testicular histology; drakes

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Introduction

L-carnitine, with extensive physiological roles, is an essential nutrient for the health. It is concentrated in the epididymis and sperm. Despite blood-testis barrier, carnitine is also highly concentrated in testis. It plays an important role not only in initiating sperm motility, promoting sperm maturation and enhancing sperm fertilizing, but also regulates Sertoli cell functions and protects sperms against oxidative damage, reduces apoptosis of spermatogenic cells and inhibits sperms aggregation (Abdelrazik and Agrawal, 2009). L-carnitine (β-hydroxyl-γ-N-trimethylamino butyrate) exists naturally in micro-organisms, plants and animals and is required for the long chain fatty acid transfer from cytoplasm to mitochondrial matrix for subsequent β-oxidation and energy production (Eder, 2000). Although cereal grains and their by-products have a low L-carnitine content (Baumgartner and Blum, 1997), they usually represent the major component of poultry diets. Consequently, L-carnitine supplementation in diet or in drinking water would be useful for facilitating fatty acid oxidation. L-carnitine is a conditional essential amino acid that plays an important role as a cofactor in cellular energy production in the mitochondrial matrix. L-carnitine aids in the transport of activated acyl groups across the mitochondrial inner membrane, and it is needed for the oxidation of long-chain fatty acids in the mitochondria of all cells (Ferrari et al., 2004). L-carnitine is a natural, vitamin-like amino-acid, synthesised within the body from lysine and methionine (Vaz and Wanders, 2002), and is very important in the metabolism of lipids. It carries long-chain fatty acids to the mitochondria for beta-

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oxidation, which produces energy (ATP) needed by the cells for proper functioning (Hoppel, 2003). We can find scant information in literature regarding the positive effects that L-carnitine exerts on male reproductive processes, especially on spermatogenesis. The compound increases sperm concentration and motility in men with idiopathic asthenozoospermia (Matalliotakis et al., 2000), in adult male chickens (Neuman et al., 2002), and rats (Palmero et al., 1990). An addition of L-carnitine in the ration increases its concentration in the epididymal tubules and, consequently, in the spermatozoa (Jeulin et al., 1994). Inside a sperm cell, L-carnitine transports fatty acids to the mitochondria, where they undergo beta-oxidation leading to the generation of metabolic energy needed by the sperm cells for their progressive movement (Jacyno et al., 2007). The concentration of carnitine and acetylcarnitine increases continuously during the epididymal passage when sperm motility and fertilizing ability develop (Jeulin et al., 1988). Carnitine and acetylcarnitine act as the first and second scavenger agent to remove acetyl-CoA from the cell and may replace poly unsaturated fatty acids (PUFA) in the seminal plasma membrane phospholipids (Arduini, 1992). Previous studies have shown that seminal free L-carnitine concentration correlates with sperm count and motility in humans (Bornman et al., 1989). Carnitine increases the activity and levels of antioxidant enzymes like superoxide dismutase (SOD) in aging rats (Kalaiselvi and Panneerselvam, 1998). Carnitine can also work together with SOD to preserve the lipid membrane surrounding sperm, thus reducing lipid peroxidation and protecting cells from peroxidation damage (Sikka et al., 1995). It was hypothesized that breeder birds and roosters fed diets supplemented with L-carnitine may show an improvement in semen traits and fertility parameters by preventing lipid peroxidation of sperm membranes (Sarica et al., 2007). Thus, the objective of the present study was to determine the effects of different levels of dietary L-carnitine supplementation on testicular histology in Iraqi drakes.

Materials and Methods

The main aim of the present study was to assess the effect of feeding mixture supplemented with L-carnitine on histological traits of testes in Iraqi drakes. A total of 48 Iraqi drakes, 30 weeks old, with an average weight of 1.57-2.00 kg were used in this study. They were randomly divided into 4 equal groups (each group contained 12 drakes) according to the dietary L-carnitine (L-Carnitine Xtreme 60 ct, Dymatize Nutrition Company, USA) supplementation for 12 weeks (0 mg / kg diet; control group (T_1), 50 mg/kg diet (T_2), 100 mg/kg diet (T_3) and 150 mg/kg diet (T_4). Each group was divided into three replicates of 4 drakes each. A photoperiod of 24 h was maintained. Food and water were provided *ad libitum* and the diets were presented in mash form. They were formulated to be isocaloric and isonitrogenous and their composition were determined according to the NRC (1994) as shown in Table 1. The experiment was started at the age of 32 weeks.

At the end of experiment, five males from each treatment group were randomly slaughtered to determine histological traits. The testes from all slaughtered drakes were collected, fixed in 10% buffered formalin solution, dehydrated in a graded series of ethanol and embedded in paraffin wax. Samples were sectioned at 5 µm thick on a rotary microtome, mounted on glass microscope slides with DPX and stained with haematoxylin and eosin.

Histological traits of testes including seminiferous tubules diameter, germinal cell thickness, seminiferous tubules diameter, volume density and relative weight of seminiferous tubules components (spermatogonia, spermatocytes, spermatids, sperms, total spermatogenic cells, Sertoli cells, vacuoles, lumen, basement membrane and total seminiferous tubules components), volume density and relative weight of interstitial tissue components (myoid cells, Leydig cells, blood vessels, interstitial spaces, total interstitial tissue components, and the ratio of total seminiferous tubules components to total interstitial tissue components).

To obtain volume of the testis components including seminiferous tubules and interstitial tissue, first the volume densities (Vv) of the seminiferous tubule components were determined by points counting method adopted by Al-Daraji et al. (2012). Briefly, an ocular square grid of 20 intersecting lines (with 228 test points) was superimposed on microscopic fields per tissue type with image software as shown in Figure 1. The software operates on live images that are transferred onto a 17-inch monitor. The image plug-in analyze was applied, placing a grid over the micrographs (grid function) and adjusting the number of points (area per point function). The number of intersections on the grid overlying the tissue component of interest was counted, and the ratio of these valid points (which were not located on the blank area) in the seminiferous tubules of each field were counted to the total number of the grid points that hit testis tissue in the photomicrograph and was considered to be the volume density of that component as shown in the following equation (Al-Daraji, 1998):

$$Vv = \frac{Pn}{Pt}$$

Where:

Vv: is the volume density of the tissue component of interest.

Table 1: Percentage of ingredient and calculated chemical analysis of experimental basal diet

Ingredients	(%)
Yellow corn	39
Wheat	33.7
Soya bean meal (44 %)	13
concentration protein*	5
Limestone	6
Vegetable oil	2
Dicalcium phosphate	1
NaCl	0.3
Total	100
Calculated Chemical composition**	
Crud Protein (%)	15.2
Energy (kcal/kg)	2927,3
Lysine (%)	0.7
Methionine (%)	0.3
Cysteine, %	0.25
Calcium (%)	2.7
Available Phosphorus (%)	0.3

*Concentration protein (BROCON – 5 SPECIAL W) each 1kg of vit. and min. premix contains: 3.25% Crud protein; 3.5% Crud fat; 1% Crud fiber; 6% Calcium; 3% Available phosphorus; 2.2% sodium; 3.5% methionine; 3.90% methionine + cysteine; 3.25 % Lysine; 2100 kcal/kg metabolizable energy; 200000 IU Vit. A; 40000 IU Vit.D_3; 500mg Vit. E; 30 mg Vit. K_3; 15 mg Vit. B_1, B_2; 150 mg Vit. B_3; 20 mg Vit. B_6; 300 mg Vit. B_{12}; 10 mg Folic acid; 50 mg Biotin; 800 mg Zinc; 100 mg Copper; 15 mg Iodine ; 1 mg Iron; 2 mg Selenium; 1.2mg Manganese; 6 mg Cobalt; and antioxidant 90 mg; **Calculated Chemical composition analysis adopted by NRC (1994).

Pn: is the number of intersections on the grid overlying the tissue component of interest.

Pt: is the total number of points on the grid (228).

The weight of seminiferous tubules components in the testis could be calculated by multiplying this volume density of each component by the relative weight of the testis (Al-Daraji, 1998).

The following components were evaluated in the testes of both control and treatments groups: the average number of the cells of the spermatogenic series (spermatogonia, spermatocyte, spermatid and spermatozoa) along with Sertoli cells in seminiferous tubules. The numbers of spermatogenic cells from a total of 3 random areas were counted to obtain the mean numbers of spermatogenic cells. The numbers of lumen, vacuoles, basement membrane, and total seminferous tubules components were also counted in the same slide as it is represented in Figure 2.

Sections were also analyzed morphometrically by using the same procedure to determine the volume densities (Vv) of the components of the interstitial tissue. Analysis was consisted of determination of the volume densities of Leydig cells, interstitial spaces, myoid cells, blood vessels and total interstitum (Fig. 3). However, the relative weight of each component of interstitial tissue was also determined by multiply the volume density of each component of interstitial tissue by relative weight of testis (Al-Daraji, 1998).

By using the same slide, the measurements of dimensions of seminiferous tubules were also determined. These measurements included semininferous tubules diameter, germinal cell thickness and semininferous tubules lumnen diameter. The measurement of the seminiferous tubule diameter (S.D) was taken twice on each slide. The tubular diameter was calculated as the average of the two measurements across the minor and major axes. However, mean diameter of seminiferous tubules lumen diameter (L.D) and germinal cells thickness (G.T) were also measured by using a special program designed for this purpose (Al-Daraji et al., 2012) by determining the distance between the two points to be measured and then choose the required measurement unit, as the program allows the measurement of the different units of measurement dimensions and as needed (Fig. 4).

The data were analyzed using an analysis of variance (ANOVA) with the general linear model procedure of the SAS program (SAS Institute, 2004). The means of variables were compared using Duncan's multiple- range test (Duncan, 1955).

Results

As shown in Table 2, the administration of L-carnitine at levels of 50, 100 or 150 mg/Kg of diet for three months resulted in a significant increase ($P \leq 0.01$) in the absolute and relative weight of testes, germinal cell thickness, seminiferous tubules lumen diameter and seminiferous tubules diameter compared with the control group.

Supplemental dietary carnitine significantly ($P \leq 0.01$) increased the volume density of spermatocytes, spermatids, sperms, total spermatogenic cells, Sertoli cells and total no of seminiferous tubules components and significantly ($P \leq 0.01$) decreased the volume density of vacuoles and lumen. However, the supplementation of dietary L-carnitine at levels of 50, 100 or 150 mg/kg to a basal diet did not influence ($P \geq 0.01$) volume density of spermatogonia and basemen membrane in drakes (Table 3).

L-carnitine also had a significantly ($P \leq 0.01$) positive effect in terms of relative weight of spermatogonia, spermatocytes, spermatids, sperms, total spermatogenic cells, Sertoli cells, total seminiferous tubules (Table 4).

Interstitial tissue components assay showed a highly significant ($P \leq 0.01$) increase in volume density of Leydig cells, blood vessels and total interstitum and a significant decrease in the volume density of interstitial spaces and total seminiferous tubules componenets/total interstitum componenets ratio in L-carnitine treated groups compared to the control group.

Table 2: Effect of dietary L-carnitine on tests weight and seminiferous tubules measurements of drakes

Traits	Treatments			
	T1	T2	T3	T4
Absolute tests weight (g)	14.73±0.86^{c}	17.87±1.09^{b}	19.14±1.12^{b}	24.65±1.25^{a}
Relative testes weight (%)	0.78±0.08^{c}	0.89±0.01^{b}	0.98±0.01^{b}	1.06±0.03^{a}
Germinal cell thickness (µm)	82.73±2.94^{c}	91.44±3.17^{b}	93.27±3.07^{b}	98.46±4.39^{a}
Seminiferous tubules lumen diameter (µm)	148.30±5.94^{c}	160.52±5.61^{b}	172.59±5.33^{a}	178.48±6.41^{a}
Seminiferous Tubules diameter (µm)	213.76±11.58^{c}	343.40±9.63^{b}	359.13±10.08^{b}	375.40±8.42^{a}

T_1: Control, T_2: 50 mg/kg of diet, T_3: 100 mg/kg of diet, T_4: 150 mg/kg of diet; Means in same row with different superscripts differ significantly (P≤0.01)

Table 3: Effect of dietary L-carnitine on volume density (%) of seminiferous tubules components of drakes testes

Traits	Treatments			
	T1	T2	T3	T4
Spematogonia	16.56±1.04	15.09±0.65	14.28±1.19	12.74±1.66
Spermatocytes	14.65±0.41^{b}	17.50±0.65^{a}	20.28±0.65^{a}	20.16±0.85^{a}
Spermatids	16.72±0.75^{b}	19.91±1.09^{a}	22.30±1.22^{a}	19.61±1.08^{a}
Sperms	4.27±0.51^{b}	7.33±0.69^{a}	11.29±0.93^{a}	12.04±1.37^{a}
Spermatogenic cells	52.11±1.54^{c}	59.83±1.39^{b}	68.15±1.13^{a}	64.55±1.5^{a}
Sertoli cells	15.29±0.82^{b}	18.25±0.48^{a}	19.00±0.41^{a}	20.58±0.22^{a}
Vacuoles	2.99±0.41^{a}	2.78±0.91^{b}	1.31±0.36^{b}	1.35±0.61^{b}
Lumen	15.41±1.74^{a}	11.75±0.82^{b}	10.71±0.51^{b}	8.13±0.29^{b}
Basement membrane	3.74±0.95	3.98±0.83	3.22±0.88	3.64±0.86
Total seminiferous tubules	89.54±0.74^{b}	96.59±1.09^{a}	102.39±2.44^{a}	98.25±2.69^{a}

T_1: Control, T_2: 50 mg L-carnitine /kg of diet, T_3: 100 mg L-carnitine /kg of diet, T_4: 150 mg L-Carnitine /kg of diet; Means in same row with different superscripts were significantly different (P≤0.01)

Table 4: Effect of dietary L-carnitine on relative weight (%) of seminiferous tubules components of drakes testes

Traits	Treatments			
	T1	T2	T3	T4
Spematogonia	12.92±0.08^{c}	13.43±0.01^{b}	13.99±0.01^{a}	13.50±0.05^{a}
Spermatocytes	11.43±0.03^{c}	15.58±0.01^{b}	19.87±0.01^{a}	21.37±0.03^{a}
Spermatids	13.04±0.06^{b}	17.72±0.01^{a}	21.85±0.01^{a}	20.79±0.03^{a}
Sperms	3.33±0.04^{c}	6.52±0.01^{b}	11.06±0.01^{a}	12.76±0.04^{a}
Spermatogenic cells	40.72±0.22^{c}	53.25±0.03^{b}	66.79±0.04^{a}	68.42±0.15^{a}
Sertoli cells	11.93±0.07^{c}	16.24±0.00^{b}	18.62±0.00^{b}	21.81±0.01^{a}
Vacuoles	2.33±0.03	2.47±0.01	1.28±0.00	1.43±0.02
Lumen	12.02±0.14	10.46±0.01	10.50±0.01	8.62±0.01
Basement membrane	2.92±0.08	3.54±0.01	3.16±0.00	3.86±0.01
Total of seminiferous tubules components	69.92±0.31^{c}	85.96±0.03^{b}	100.35±0.02^{a}	104.14±0.04^{a}

T_1: Control, T_2: 50 mg L-carnitine /kg of diet, T_3: 100 mg L-carnitine /kg of diet, T_4: 150 mg L-Carnitine /kg of diet; Means in same row with different superscripts were significantly different (P≤0.01)

Table 5: Effect of dietary L-carnitine on volume density (%) of interstitial tissue components of drakes testes

Traits	Treatments				Level of significance
	T1	T2	T3	T4	
Myoid cells	1.55±0.14	1.64±0.17	1.68±0.18	1.79±0.14	N.S
Leydig cells	10.04±0.91^{b}	14.25±0.69^{c}	15.96±0.89^{a}	18.37±0.82^{a}	**
blood vessels	0.20±0.07^{b}	0.58±0.08^{a}	0.49±0.08^{a}	0.61±0.10^{a}	*
Interstitial spaces	4.01±1.72^{a}	3.43±0.72^{b}	3.01±0.14^{b}	2.12±0.12^{c}	*
Total interstitum	15.8±2.84^{c}	19.9±1.66^{b}	21.14±1.29^{a}	22.89±1.18^{a}	**
Total seminiferous tubules/ total interstitum	5.66±0.26^{a}	4.85±0.65^{b}	4.84±1.89^{b}	4.29±2.27^{c}	*

T_1: Control, T_2: 50 mg L-carnitine /kg of diet, T_3: 100 mg L-carnitine /kg of diet, T_4: 150 mg L-Carnitine /kg of diet; Means in same row with different superscripts were significantly different; N.S: Non-significant; *: (P<0.05); **: (P≤0.01).

Table 6: Effect of dietary L-carnitine on relative weight (%) of interstitial tissue components of drakes testes

Traits	Treatment				Level of significance
	T1	T2	T3	T4	
Myoid cells	1.21±0.01^{c}	1.46±0.00^{b}	1.65±0.00^{a}	1.90±0.00^{a}	*
Leydig cells	7.83±0.07^{b}	12.68±0.01^{a}	15.64±0.01^{a}	19.47±0.02^{a}	**
blood vessels	0.16±0.01^{c}	0.52±0.00^{b}	0.48±0.00^{b}	0.65±0.00^{a}	**
Interstitial space	3.13±0.14	3.05±0.01	2.95±0.00	2.25±0.00	N.S
Total interstitum	12.32±0.23^{d}	17.71±0.02^{c}	20.72±0.01^{b}	24.26±0.04^{a}	**
Total sominiferous tubules/ total interstitum	2.37±1.35^{a}	1.85±1.50^{b}	1.62±2.00^{c}	1.47±1.00^{c}	*

T_1: Control, T_2: 50 mg L-carnitine/kg of diet, T_3: 100 mg L-carnitine/kg of diet, T_4: 150 mg L-Carnitine/kg of diet; Means in same row with different superscripts were significantly different; N.S: Non-significant; *: (P<0.05); **: (P≤0.01).

Furthermore, no significant difference in volume density of myoid cells between experimental groups was found (Table 5).

Results also revealed the supplementing the diet of drakes with different levels of L-carnitine resulted in significant increase in relative weight of myoid cells, Leydig cells, blood vessels and total interstitum and significant decrease in relative weight of total seminiferous tubules/total interstitum, while there was no significant difference between all experimental groups concerning the relative weight of interstitial spaces (Table 6).

Discussion

The results of our experiment have demonstrated a positive effect of L-carnitine on the testis weight, seminiferous tubules measurements and volume density and relative weight of the active components of seminferous tubules and interstitial tissue. These results may be due to the beneficial effects of L-Carnitine supplementation. Lipids are a basic component of semen, contributing to the membrane structure of spermatozoa, the metabolism of the sperm cells, and their ability to fertilize the female gamete. There is considerable evidence to indicate that the lipid composition of the sperm membrane is a major determinant of the cold sensitivity, motility, and overall viability of spermatozoa (Kelso et al., 1997). The presence of the high concentrations of long-chain PUFA of the n-6 series in avian spermatozoa increases their susceptibility (vulnerability) to lipid peroxidation and limits the viability of chicken and turkey spermatozoa. Lipid peroxidation plays a key role in the aging of spermatozoa by shorting its lifetime *in vivo* as well as during the *in vitro* conservation of sperm for artificial insemination. The peroxidation process comes with extensive structural alterations, especially in the acrosomal section of the spermatozoa, fast and irreversible loss of motility, extensive metabolic changes, and high rate of leakage of intracellular spermatic constituents. Semen or spermatozoa is normally equipped with diverse enzymatic (glutathione peroxidase, SOD) protection against lipid peroxidation in birds. However, an increase in lipid peroxidation may be correlated with decreased antioxidant enzyme status and activities. L-Carnitine supplementation increased overall antioxidant enzyme activities as a function of the duration of treatment, thus decreasing the levels of free radicals available for lipid peroxidation (Sarica et al., 2007). The results of our previous experiment demonstrated a positive effect of L-carnitine in the chicken semen (Al-Daraji and Tahir, 2013a). Clinical studies suggest that L-carnitine supplementation over a period of 3 to 6 months can positively affect sperm concentration, sperm count, percentage of motile sperm, and the percentage of sperm with rapid progression in human (Lenzi et al., 2003). L-carnitine has also antioxidant properties that protect sperm membranes against toxic reactive oxygen species. Pignatelli et al. (2003) demonstrated that carnitine reduces oxidative stress via interference with arachidonic acid incorporation into phospholipids and protein kinase C mediated NADPH oxidase system. It is also proposed that carnitine exerts antioxidant properties as a result of repairing mechanism by which elevated intracellular toxic acetyl-CoA is removed and fatty acids in membrane phospholipids are replaced (Vicari and Calogero, 2001). Ahmed et al. (2011) found that L-carnitine level in seminal plasma plays an essential role in maintaining male fertility. A significant positive correlation was detected between L-carnitine concentrations and the number and motility of spermatozoa (Al-Daraji and Tahir, 2013b). When these observations are considered along with the well-established role L-carnitine in sperm energy production, maturation and antioxidant properties, it creates a rationale for treatment with L-carnitine in many cases of male infertility.

Previous studies were performed to evaluate the role of L-carnitine in spermatogenesis and seminal fluid. In an attempt to characterize the protective action of Acetyl L-carnitine (ALC) using an *in vivo* system, the recovery and maturation process of mouse spermatogenesis was investigated. Mice were exposed to irradiation to deplete the spermatogonia and then were given ALC. The sperm population in the mice that received ALC demonstrated a quicker recovery

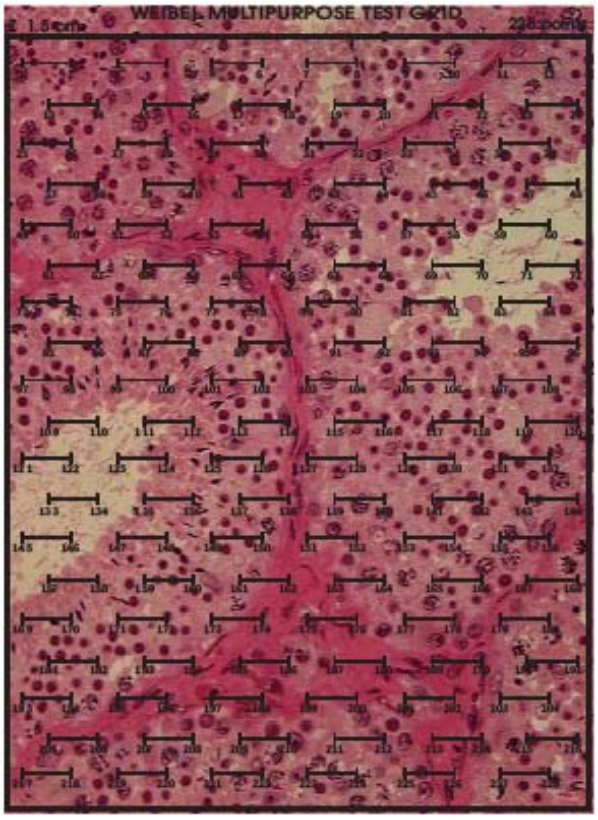

Fig. 1: Photomicrograph of drakes testicular tissue with a 228-point overlay grid generated by plug-in of image software

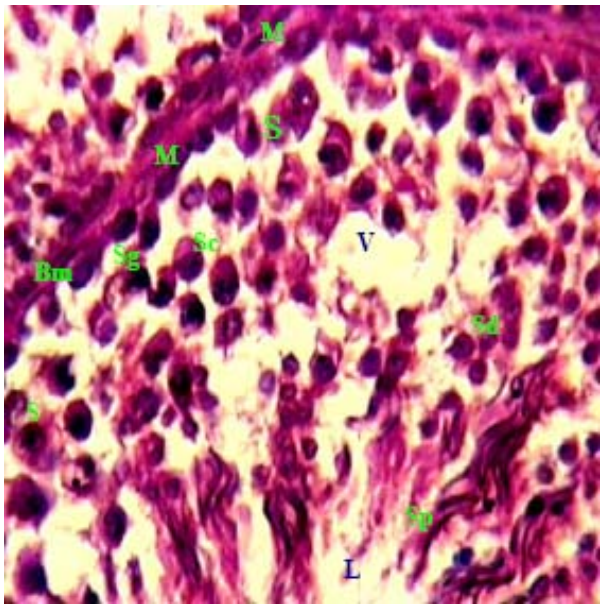

Fig. 2: Histological sections of the seminiferous tubules of testes. S = Sertoli cells, V = vacuoles, Sg = spermatogonia, Bm = basement membrane, M = myoid cells, Sc = spermatocytes; Sd = spermatid, and Sp = sperm; × 400.

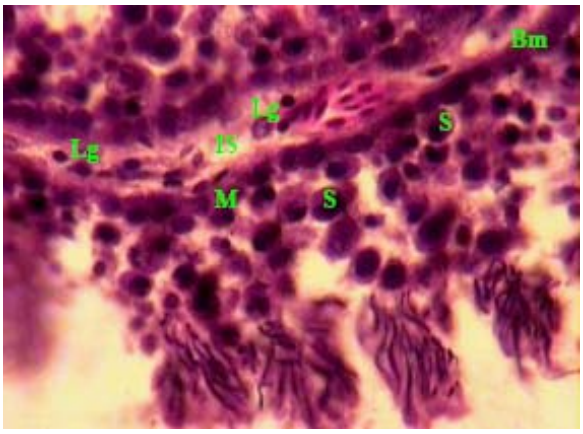

Fig. 3: Histological sections of the seminiferous tubules of testes. S = Sertoli cells, M= myoid cells, Lg= Leydig cells, and IS= Interstitial spaces; × 400.

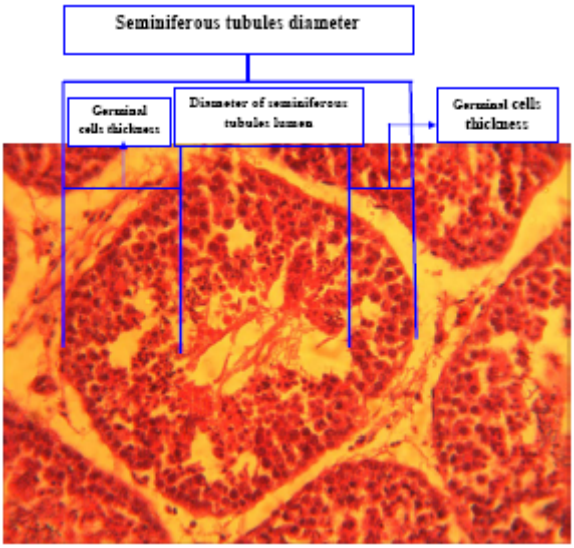

Fig. 4: Measurements of dimensions of the seminiferous tubles; × 100

throughout the maturation process than the spermatozoa in those that did not receive ALC (Al-Rubiey, 2012). Therefore, it appears that ALC could influence the early stages of spermatogenesis with consequent favourable effects on DNA repair and on proliferation of regenerating germ cells (Al-Rubiey, 2012). L-carnitine plays a key role in sperm metabolism by providing readily available energy for use by spermatozoa, which positively affects sperm motility, maturation and the spermatogenic process. This beneficial effect is mediated by the transport of long chain fatty acids across the inner membrane of the mitochondria for utilization in metabolism through β-oxidation (Matalliotakis et al., 2000). An ample concentration 0f carnitine has been detected in the rat testicle (half of the epididymal concentration), which suggests that it plays

a role at the testicular level. In support, high concentration of acetyl L-carnitine transferase has been detected in primary spermatocytes and developing testicular tissue (Aliabadi et al., 2012). Carnitine may affect testicular sperm maturation indirectly via the stimulation of Sertoli cells glucose uptake. In general, Sertoli cells represent a very important site for the control of the spermatogenic process (Bielli et al., 2001). The addition of L-carnitine to Sertoli cell cultures resulted in a considerable increase in pyruvate and lactate secretion, which are known to represent essential energy substrates for germ cell maturation (Palmero et al., 2000). Al-Rubiey et al. (2012) found significant increase in the number of Leydig cells ($P<0.05$) in groups of rats treated with L-carnitine as compared to placebo. A potential important biomarker of testicular function is the active carnitine acetyltransferase, which is contained within the spermatozoa. The activity of this enzyme is 7-fold higher in the diplotene primary spermatocytes than in the spermatogonia, indicating that carnitine acetytransferase may be useful as a marker enzyme of germ cell differentiation in the testis (Vernon et al., 1971). It appears that carnitine could influence the early stages of spermatogenesis with consequent favourable effects on DNA repair and on proliferation of regenerating germ cells (Amendola et al., 1991).

Conclusion

In our study, administration of L-carnitin to diet of drakes at 0, 50, 100 and 150 mg/kg led to significant improvement in testis weight, measurements of seminiferous tubules and volume density and relative weight of active components of seminiferous tubules and interstitial tissue. Therefore, this study suggests that the administration of L-carnitine may be beneficial for improving male sexual performance.

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